Commentary

The art of killing

Double stroke with apoptin and survivin as a novel approach in cancer therapy

Soumya Panigrahi,1,2,* Thomas Klonisch3 and Marek Los4

1Department of Physiology; University of Manitoba; Winnipeg, Canada; 2Manitoba Institute of Cell Biology; Cancer Care Manitoba; Winnipeg, Canada; 3Department of Human Anatomy and Cell Science; University Manitoba; Winnipeg, Canada; 4BioApplications Enterprises; Winnipeg, Manitoba Canada

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Despite the introduction of first cancer chemotherapies in the early 20s of the last century cancer still remains the second most common cause of death in developed countries. Several anticancer drugs have been introduced to the clinic, and at last count, a few dozen more are at various stages of development. With the accumulation of knowledge about the biology of cancer in recent years, more researchers now aim at targeting signaling pathways that are frequently upregulated in certain types of cancer. These so-called targeted therapies, although not always effective as a single-agent treatment, have generally low to negligible side effects.1,2 In addition to therapeutics with known molecular targets, experimental drugs have emerged which are more or less selectively toxic against cancer cells but spare normal, healthy cells. However, their exact molecular mechanisms of action are mostly unclear. This group includes viral proteins R4orf4, apoptin, human cytokine TRIAL, and the human lipoprotein complex, HAMLET.3,4

In the current issue of Cancer Biology & Therapy, Liu and coauthors aim to develop a new anticancer therapy by combined delivery of apoptin and micro-RNA-based inhibition of survivin.5 Both therapeutic modalities are incorporated into a single delivery vector. This approach assures temporary- and spatially-synchronised expression and action of both therapeutic agents.

Survivin is an atypical member of the ‘inhibitor of apoptosis protein’ (IAP) family of proteins that inhibits both the activation and activity of already mobilized caspases, a family of proteases involved primarily in the propagation and execution of apoptotic cell death. Unlike other IAP-family members, survivin’s action is more complex. Survivin expression is upregulated in human cancers. It is associated with chemo- and radiotherapy resistance, and linked to poor prognosis. Survivin’s expression is highest in the late G2 and the M-phase of the cell cycle and it appears to function both as apoptosis inhibitor and cell cycle regulator.6,7 With the onset of mitosis, survivin binds to microtubules of the mitotic spindle apparatus and both the cyclin-dependent-kinase inhibitor p21Cip1/Waf1 and caspase-3 colocalize with survivin at centrosomes. Inhibition of survivin function leads to caspase-3 activation and apoptotic cell death. Moreover, it also causes defects in cell division that manifest as aneuploidy, multinucleation, and supernumerary centrosomes.8 Previous experiments that involved targeting survivin’s expression with specific ribozymes or with antisense nucleotides induced apoptosis in various cell lines or broke resistance to conventional anticancer drugs.6 The dual mode of survivin’s action has attracted attention of the pharmaceutical industry. For example, Isis Pharmaceuticals and Abbot Laboratories are testing antisense-based approaches that interfere with the expression of survivin.9,10

Apoptin gained significant attention as a selective killer of cancer cells, and thus it may not only serve as a lead for the development of novel drugs, but also as a tool to delineate the critical molecular differences between the normal cellular state and neoplasia. Apoptin is a small (121 amino acids long, 14 kDa) basic protein encoded by the third open reading frame (VP3) of chicken anemia virus.3 Its selective anticancer properties were discovered by the group of Noteborn, in 90s.11 Its cancer-selective mode of action, although still not fully understood, appears to involve multiple mechanisms.

Apoptin selectively kills both p53-positive and negative cancer cells.3,12 The mitochondrial/apoptosome-dependent death pathway mediates apoptin induced cell death.13-15 Apoptin induced cell death requires PI3-K activation, nuclear transfer of Akt, and cytoplasmic transfer of phosphorylated Nur77.15-17 Apoptin interacts with both p85α and Akt via its proline-rich sequence.16,18 It appears that apoptin is able to “hijack” these cell survival pathways and redirect them to fuel apoptotic cell death. In normal cells, apoptin localizes to the cytoplasm, whereas in cancer cells it is predominantly found in the nucleus. The preferred nuclear localization of apoptin in malignant cells is governed by the combined actions of an activated nuclear localization signal and the suppression of a nuclear export signal.19 Phosphorylation of Thr-108 at apoptin’s C-terminus appears to play an important role in the above events. Apoptin interacts with the components of anaphase-promoting complex and interferes with cell division.20 Exhibiting basic properties (pK ~10.6), apoptin will interact directly with heterochromatin and with DNA ends, thus, it may interfere with gene transcription, DNA synthesis and DNA repair.21

The choice of combining apoptin expression with mi-RNA-based surviving inhibition for cancer therapy is brilliant as both molecules have diverse, multiple, yet partly overlapping targets in the cell. It is
safe to assume that the interference with survivin expression will facilitate apoptin's action in many ways. (i) Apoptin-induced cell death involves caspases, thus, the absence of survivin will facilitate their active state and ensure completion of the apoptotic process. (ii) Among other factors, survivin expression is regulated by the PI3-K/Akt pathway. The interference of apoptin with these pathways may affect survivin expression and amplify apoptin's action. (iii) Apoptin interacts with the anaphase-promoting complex. The lack of survivin's action as a guardian of mitotic spindle formation will facilitate apoptin-triggered cell death. These are major effects that likely occur as a result of survivin knock-down in the presence of apoptin when applied simultaneously in cancer therapy. The molecular mechanisms of action of both survivin and apoptin are currently the subject of intense investigation and additional molecular events triggered by this combined therapeutic strategy are likely to be discovered.

Proper targeting is a major issue when new gene therapies are designed. Although its suitability in animal models remains to be tested, the combined and simultaneous therapeutic system of miRNA-based specific knockdown of survivin, and overexpression of apoptin described by Liu and colleagues has built-in selective advantages: (a) survivin is predominantly (if not exclusively) expressed in rapidly dividing cells, and (b) apoptin selectively kills cancer cells. The therapy however may carry potential complications that need to be carefully experimentally addressed before this vector approach is to be moved into clinical trials. For example, the inhibition of survivin expression may sensitize also normal cells to apoptin, and apoptin's interference with the cell cycle may cause survivin to induce aneuploidy and tumorigenic dedifferentiation of normal cells.

References


Figure 1. AUTHOR: please provide figure legend.